Prex2 Cas9-CKO Strategy

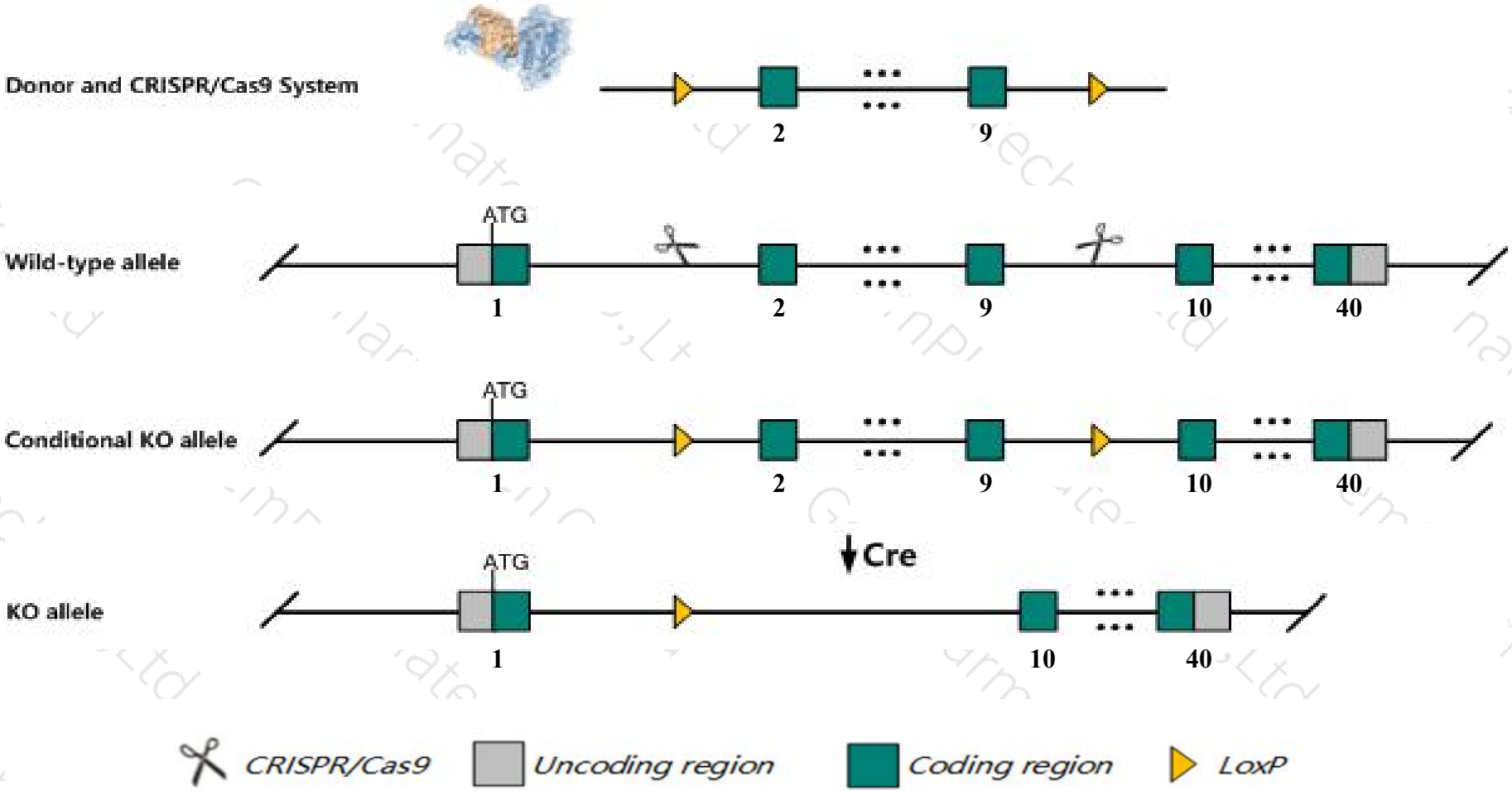
Designer:Xueting Zhang
Reviwer:Yanhua Shen
Date:2020-02-13

Project Overview

Project Name	***Prex2***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Prex2* gene. The schematic diagram is as follows:



Technical routes

- The *Prex2* gene has 8 transcripts. According to the structure of *Prex2* gene, exon2-exon9 of *Prex2-201* (ENSMUST00000027056.11) transcript is recommended as the knockout region.The region contains 952bp coding sequence. Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Prex2* gene. The brief process is as follows:CRISPR/Cas9 system and Donor were microinjected into the fertilized eggs of C57BL/6JGpt mice.Fertilized eggs were transplanted to obtain positive F0 mice which were confirmed by PCR and sequencing. A stable F1 generation mouse model was obtained by mating positive F0 generation mice with C57BL/6JGpt mice.

- The flox mice will be knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues and cell types.

Notice

- According to the existing MGI data,Mice homozygous for gene trapped alleles may exhibit abnormal Purkinje cell dendrite morphology, a mild motor coordination defect that progressively worsens with age, hypoactivity, impaired glucose tolerance and/or insulin resistance.
- Transcript *Prex2*-202&203&204&205&206&208 may not be affected.
- The *Prex2* gene is located on the Chr1. If the knockout mice are crossed with other mice strains to obtain double gene positive homozygous mouse offspring, please avoid the two genes on the same chromosome.
- This Strategy is designed based on genetic information in existing databases.Due to the complexity of biological processes, all risk of loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information （NCBI）

集萃药康 GemPharmatech

Prex2 phosphatidylinositol-3,4,5-trisphosphate-dependent Rac exchange factor 2 [*Mus musculus* (house mouse)]

Gene ID: 109294, updated on 12-Aug-2019

Summary

Official Symbol	Prex2 provided by MGI
Official Full Name	phosphatidylinositol-3,4,5-trisphosphate-dependent Rac exchange factor 2 provided by MGI
Primary source	MGI:MGI:1923385
See related	Ensembl:ENSMUSG00000048960
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	Depdc2; P-Rex2; AI316880; AI553603; D430013K02; 6230420N16Rik; C030045D06Rik
Expression	Broad expression in lung adult (RPKM 9.2), bladder adult (RPKM 4.1) and 17 other tissues See more
Orthologs	human all

Genomic context

Location: 1; 1 A2

See Prex2 in Genome Data Viewer

Exon count: 41

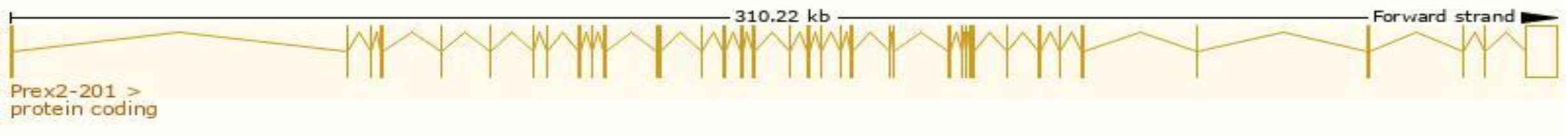
Annotation release	Status	Assembly	Chr	Location
108	current	GRCm38.p6 (GCF_000001635.26)	1	NC_000067.6 (10993273..11303683)
Build 37.2	previous assembly	MGSCv37 (GCF_000001635.18)	1	NC_000067.5 (10983546..11293763)

Transcript information（Ensembl）

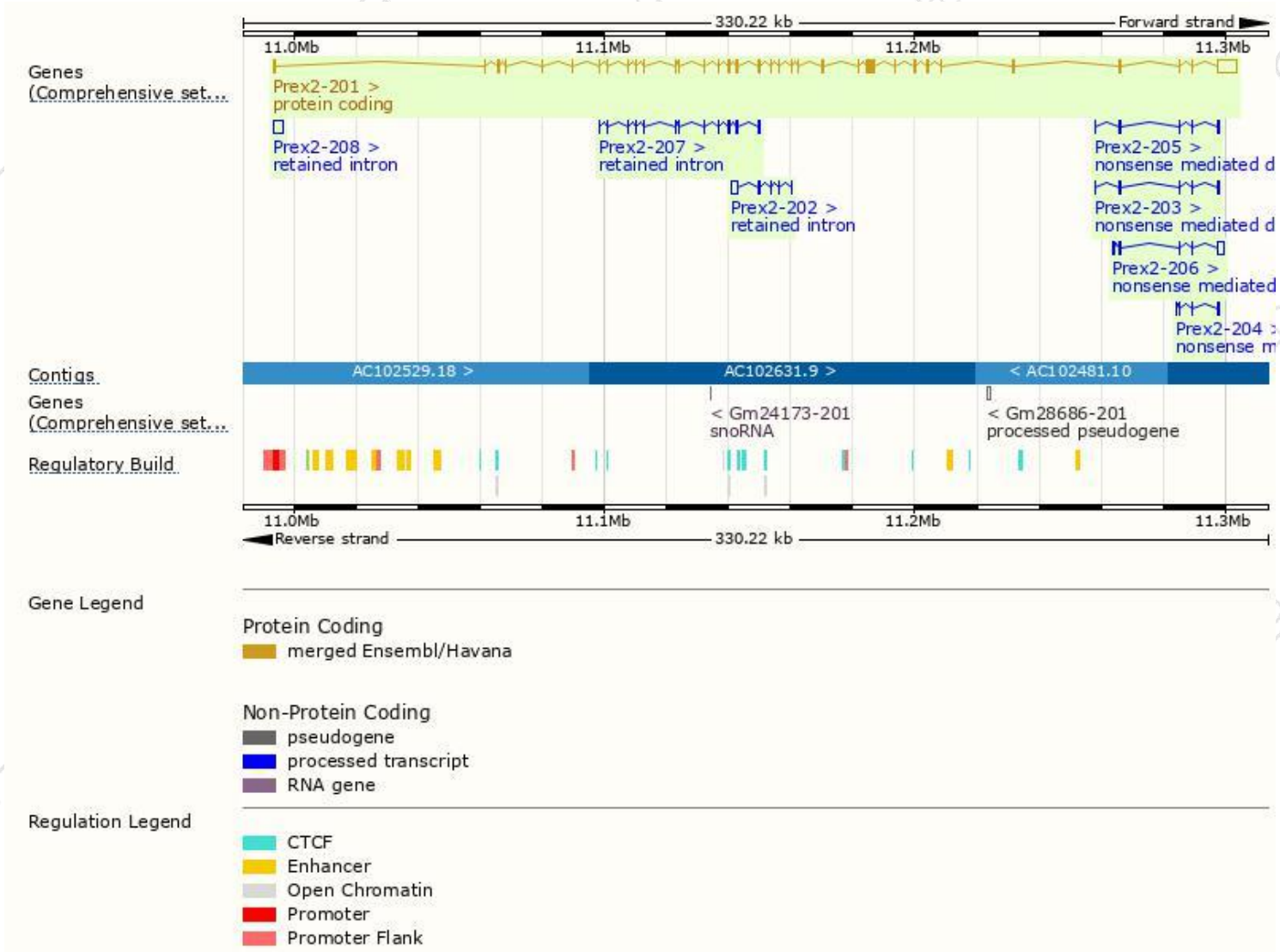
The gene has 8 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Prex2-201	ENSMUST00000027056.11	11053	1598aa	Protein coding	CCDS48217	Q3LAC4	TSL:1 GENCODE basic APPRIS P1
Prex2-206	ENSMUST00000189385.6	2449	24aa	Nonsense mediated decay	-	A0A087WS21	CDS 5' incomplete TSL:1
Prex2-204	ENSMUST00000188154.1	843	40aa	Nonsense mediated decay	-	A0A087WRD8	CDS 5' incomplete TSL:3
Prex2-203	ENSMUST00000187745.6	769	50aa	Nonsense mediated decay	-	A0A087WRV5	CDS 5' incomplete TSL:5
Prex2-205	ENSMUST00000188189.6	726	15aa	Nonsense mediated decay	-	A0A087WRG8	CDS 5' incomplete TSL:5
Prex2-208	ENSMUST00000190935.1	3084	No protein	Retained intron	-	-	TSL:NA
Prex2-202	ENSMUST00000187694.1	2949	No protein	Retained intron	-	-	TSL:1
Prex2-207	ENSMUST00000189822.1	1517	No protein	Retained intron	-	-	TSL:1

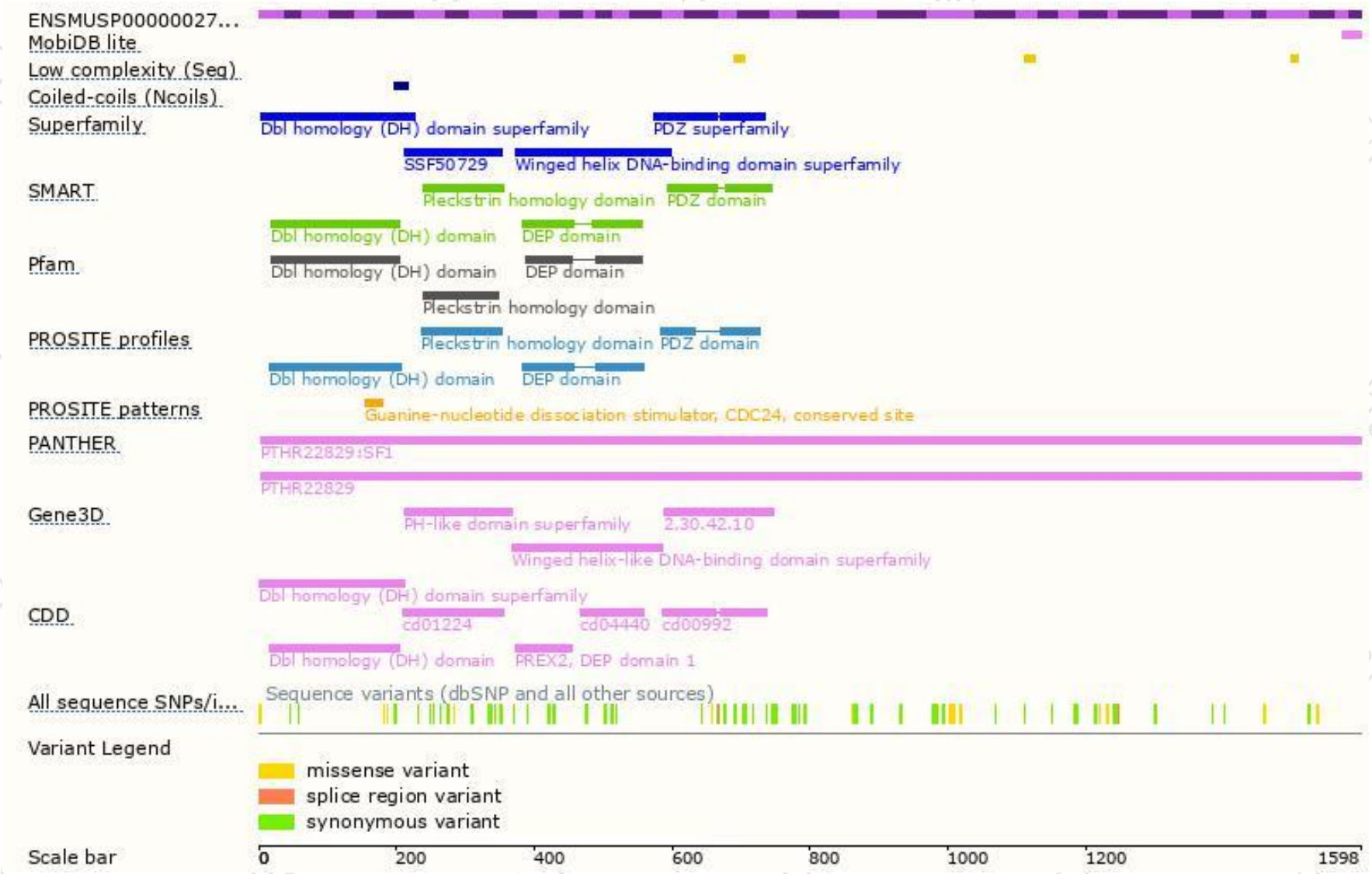
The strategy is based on the design of *Prex2-201* transcript,The transcription is shown below



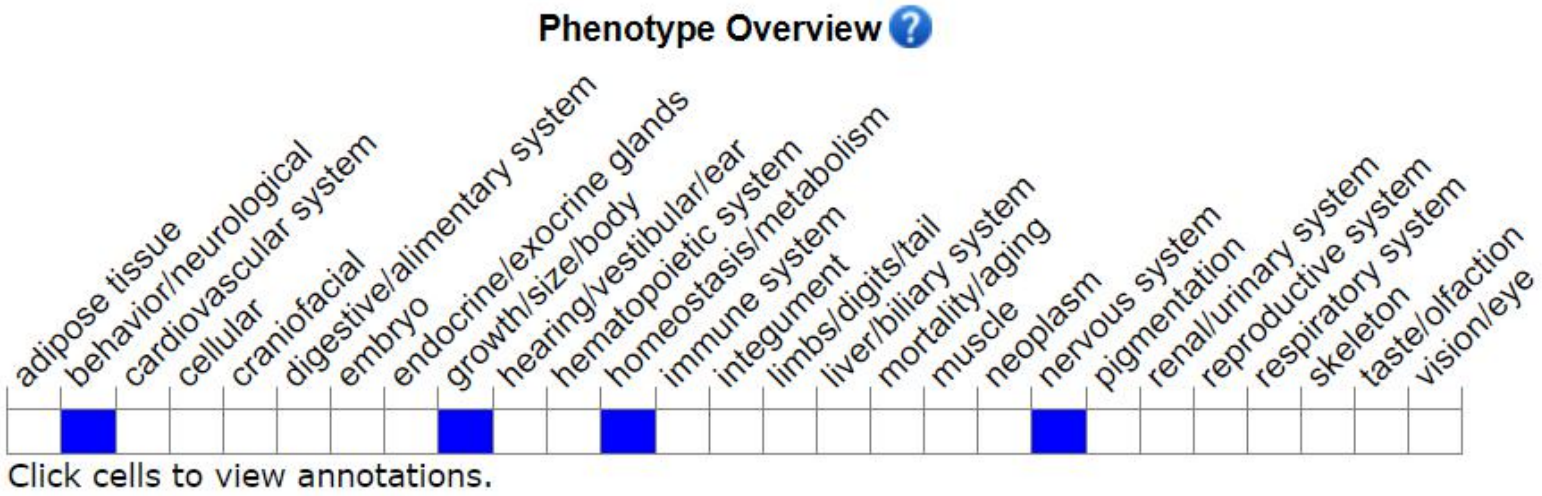
Genomic location distribution



Protein domain



Mouse phenotype description(MGI)



Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).

According to the existing MGI data,Mice homozygous for gene trapped alleles may exhibit abnormal Purkinje cell dendrite morphology, a mild motor coordination defect that progressively worsens with age, hypoactivity, impaired glucose tolerance and/or insulin resistance.

集萃药康
GemPharmatech

If you have any questions, you are welcome to inquire.

Tel: 400-9660890

集萃药康生物科技
GemPharmatech Co.,Ltd